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APPLE ROTS IN ILLINOIS.

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During the past few years considerable complaint has been made in this state of the loss of apples through rotting. The trouble has been attributed chiefly to the bitter rot fungus which, apparently, has been unusually injurious in the United States and especially in certain parts of the Mississippi Valley during this period. Beginning the past season the Experiment Station has undertaken an especial study of this and other apple rots of Illinois both in the field with reference to their distribution, damage, and prevention and in the laboratory with reference to the life history of the fungi that cause them. This bulletin includes the studies, especially of the bitter rot fungus, made from the latter point of view during the months of July, August, and September.

Bitter rot, while the chief, is not the only serious rot of apples in Illinois. A superficial study only has been made of these other rots, the most common of which are briefly mentioned here.

FRUIT BURN. As indicated by the name this is a scalding of apples due to the sun's heat. Ordinarily this trouble is confined to the fruit after it has fallen to the ground, though during the past unfavorable apple season there has been some complaint of the fruit burning on the trees. The apples as they lie on the ground begin to scorch on the south or sunny side and the brown discoloration gradually increases in size both by spreading over the surface and by extending inward, eventually forming a sort of baked apple. In its earlier stage diseased tissue taken from the interior of such apples for cultural examination gives no growth of bacteria or fungi, but in time, no doubt, these agents often aid in the destruction of the tissues. The trouble, apparently, is not a very serious one. Plate A, Fig. 1, shows an apple affected with fruit burn in its earlier stage.

BROWN ROT, *Monilia fructigena*, Pers. This rot was not seen very often the past season, though in some years it appears as a rather common cause of the rotting of summer varieties. It soon produces a complete rotting of the infected apples which turn a brown color and show the presence of the fungus by the breaking out of small, greyish, dense pustules of spore bearing mycelium. These fruiting sori are usually found only when the fruit is in a moist place, especially when it has fallen on the damp earth, though they are sometimes seen on apples hanging on the tree. Ordinarily this rot appears only on ripening summer varieties and most commonly after they have fallen from the tree. The same fungus also causes the common rotting of peaches, plums, and the occasional rotting of cherries, etc., and on some of these hosts often causes serious loss.

SOFT ROT, *Rhizopus nigricans*, Ehr. This rot does not commonly attack apples on the trees though it was so found in a fruiting condition the past season on ripe summer varieties. It is a rather soft rot which is not otherwise very characteristic unless, as rarely happens except under moist conditions, the fungus breaks out on the surface in its fruiting stage, which consists of short, somewhat bunched threads bearing on their ends the small black spore cases. The fungus is primarily more of a saprophytic than a parasitic fungus and is a common cause of the rotting of various mature vegetables and fruits. Rots like those shown in Plate A, Fig. 2, are sometimes caused by this fungus.

FRUIT BLOTCH, *Phyllosticta*, sp. What appears to be an undescribed fungous trouble of apples was first found by Professor Burrill during July of the past season in a number of places in southern Illinois. Later it was seen by the writer on a single

variety in the University orchard and it has been reported since by various persons as common on certain varieties in different parts of southern Illinois. Mr. A. A. Hinkley, of DuBois, reported it as a common trouble in his orchard during the past season and also stated that he had known it for some time but had been calling it scab. It has been found on Rambo, Ben Davis, Janet, Grimes' Golden, and Winesap and undoubtedly occurs on other varieties. So far as reported Rambo and Ben Davis were the worst affected of these.

The trouble appears at first as small dark colored blotches of quite irregular shape that are more or less scattered over the surface of the apples and quite superficial. On these are often seen quite small pustules which are the conceptacles containing the spores, which are hyaline, oval to subspherical and chiefly 7--10 μ in greatest diameter. They germinate rather readily when placed in a drop of water, sending out short, septate germ-threads. Later in the season, September to October, the disease assumed a more serious aspect. The discolored spots became enlarged and more or less merged into extended and often slightly sunken areas and the diseased tissue extended inward so that in some cases the whole apple was affected with dry rot. This later injury may possibly in part have resulted from the action of some other fungi or bacteria. Plate B, Fig. 1a—b, shows action of the fungus in its earlier and (c) in its later stage.

Cultures made by taking diseased tissues from the interior of affected apples produced a characteristic dark olive green mycelium that formed patches of rather slow growth on the medium and had not after two months' development given any sign of the formation of a spore stage.

Being unable to identify the species of this fungus, specimens were sent to the veteran mycologist, J. B. Ellis, for inspection and he suggested the possibility of its being the fructigenous form of Peck's *Phyllosticta limitata*. Specimens were then sent to Peck who reported the two as distinct. This view was confirmed by the examination of specimens from the material from which Peck described *Phyllosticta limitata*, Mr. A. F. Stewart kindly furnishing these. This fungus is also distinct from *Phyllosticta pirina*, Sacc. and *Phyllosticta prunicola* (Opiz.) Sacc., which have been reported on Pirus. So far as can be ascertained the fungus seems to be a new species and it is hoped to make further investigation of it the coming year.

During the latter part of the season when one might expect the fruiting pustules to form spores more abundantly it was diffi-

cult to find any containing spores, but instead their contents were usually in an immature condition. This suggested that the fungus was beginning the development of its permanent or ascosporic stage, and it is possible this may be found eventually.

Black Rot, *Sphæropsis Malorum*, Berk. This fungus has been known to occur in Illinois at least since 1879 when Professor French of Carbondale sent specimens of rotting apples to State Botanist Peck of New York who so identified the fungus found on them. Black rot causes more or less decay of apples everywhere, and it has been a matter of some interest in the state to know how much of the damage here is to be attributed to this source and how much to bitter rot. The observations of the past season show the latter to be the much more aggressive fungus, though it does not have the wide distribution of the former, which is likely to occur to some extent in every orchard. On green apples black rot usually appears only where they have been injured in some manner, as by worms, etc., and is most commonly found on windfalls. Ordinarily it does little damage to perfect, green fruit on the trees but sometimes on ripe summer and crab apples does more or less damage, as was found in a few cases the past summer. It is also one of the chief causes of the rotting found in market apples.

Black rot is so called because of the color assumed by the infected fruit. Often at first the rotten part is brown but in time this becomes black. The color is due to the mycelium of the fungus which while at first hyaline eventually becomes dark olive, especially at the surface of the fruit. The mycelium occurs rather abundantly through the rotten tissue and is made up of septate threads often quite irregular and of considerable size. The rotting is usually well along before the fruiting stage appears as abundant but very small black pustules at the surface of the apple. These pustules are roundish conceptacles having the outer wall made up of black or often purplish tinged cells and containing the spores. The spores often ooze out from the conceptacles in small white tendrils. While at first they are colorless and simple, they eventually become deeply colored and usually uniseptate. As the rotting advances the apples become shriveled so that finally they form small, hard, irregular mummies. Plate B, Fig. 2a—d, shows something of these progressive stages.

The black rot fungus also occurs on the leaves and branches and on these parts seems to cause more serious damage than on the green fruit. On the leaves the brownish spots produced are much like those of the leaf spots or Phyllostictæ fungi but are apt to be more irregular and larger. Ordinarily the fruiting pustules are not borne

very abundantly on these spots. The old dead twigs so often found on unpruned apple trees usually bear an abundance of the fruiting pustules and while formerly it was believed that the fungus grew on these only as a saprophyte there is now evidence that it is often responsible for the death of such twigs.

Black rot is also a fungous trouble of other of the pomaceous fruits. The fact that it occurs on these different hosts and on different parts of the same host has such differences in general appearance, together with the fact that specimens show considerable difference in color and septation of spores according to their age, has caused botanists to describe this fungus under a number of different names.

Miscellaneous Rots. Besides the preceding and the bitter rot, there are a number of other fungi, also bacteria and yeasts, that have been found in rotting market apples. Sometimes these are the primary agents of the rotting and sometimes only secondary ones. In a cultural examination of tissue taken with the usual precautions from the interior of apples at the juncture of healthy and rotten areas the following results were obtained with twenty-three samples gathered from different grocery stores in Champaign and Urbana: Four developed *Rhizopus nigricans*, soft rot; four, *Sphæropsis Malorum*, black rot; five, nothing; four, bacteria or yeasts or both; one, *Gliocladium penicillioides;* one Phyllosticta sp., fruit blotch; three, mixtures of various fungi or non-fruiting forms; one, Aspergillus sp. These cultures were each made in duplicate and usually both gave the same result, thus showing that the primary cause of the rotting was usually one agent, though no doubt in the older rotten parts it was often assisted by other forms.

Besides the above species there were also found on badly rotted apples several other fungi that apparently occurred there primarily as saprophytes, no doubt often to the detriment of the primary form causing the rotting. Such forms were species of the following genera: Cephalothecium, Penicillium, Aspergillus, Mycogone, Alternaria, and Acremoniella.

Bitter Rot.

Occurrence. None of the preceding fungi compare with the bitter rot fungus in the damage that it may cause to the apple crop, since it is a parasite that is equally at home on the green or the ripe fruit. It also has a wide distribution over the world. It seems to have periods of greater and lesser destructiveness and especially during the past two or three years has developed more vigorous in southern Illinois. While no attention has been paid to it by the

Station until recently, it has been known to exist in southern Illinois at least since 1869. During the past two years it has been found in a number of places in this part of the state, the most northern reported limit being at Urbana. It seems quite probable that it occurs, at least sparingly, in central and northern Illinois, as it has been found as far north as Maine. The general information now at hand, however, indicates that it is a more active agent in regions with a climate like southern Illinois.

DAMAGE. Something of the damage that has been caused in this state is shown by the following statement of Mr. C. H. Murray of Clay City in the transactions of the Illinois Horticultural Society for 1870, page 346. Mr. Murray says concerning an orchard of a Mr. Finch: "Last year it had at least one thousand bushels of apples on and the proprietor did not get a bushel of winter apples. The bitter rot blasts them like the breath of ruin and the promise of spring ends in disappointment and decay. Many experiments have been tried to arrest this evil but so far none of them has proved efficacious. * * * * * For ten or eleven years it gave the most bounteous returns and produced wagon loads of the finest fruit. It then began to decline. The fruit commenced to speck and the evil increased until the trees are little more than an incumbrance on the ground. * * * The Vandevers all have the 'bitter rot' also the Bellflowers but the Pippins and Summer Queen have not, but have a sweet or summer rot." The interest manifested by orchardists in southern Illinois the past two seasons shows that bitter rot is still a pest that is greatly feared and for sufficient reason, since in some orchards the entire crop has been destroyed by it.

PREVENTION. This being so serious a trouble naturally attempts have been made to prevent it. More or less success has been reported by different experimenters. Some have advocated picking the infected fruit. This to be of any service must be very thorough and commenced with the very first appearance of the rot and repeated often. Old bitter rot apples lying on the ground or mummies attached to the tree are apparently the source of infection the coming year, and these especially should be destroyed.

Spraying has been tried with more or less success. To be of value it should be commenced before the first appearance of the rot and repeated from time to time until danger is past. It is especially important that it be repeated after each rainfall. The Horticultural Department of the Station is at work along this phase of the question and whatever results are obtained will be reported.

APPEARANCE OF INFECTED FRUIT. Sometimes apples begin to rot at numerous points scattered over their surfaces, as is shown in

Fig. 1, Plate C. More commonly the infection starts at one or a few places, as is shown in (a) and (b) Fig. 2, Plate C. The character of the rot varies somewhat with the variety infected and the weather conditions, but in general the original small point of infection increases to a brown rotten area a quarter to a half an inch in diameter without showing external growths of any kind. About this time, however, numerous, small, covered, usually black pustules appear at the center. These are the fruiting sori and if the conditions are favorable they soon rupture the covering cuticle and ooze out on the surface small pinkish masses of agglutinated spores. If the variety offers too great resistance to the fungus or if the weather conditions have been unfavorable—that is, too dry—the fruiting pustules may continue to be formed but with few or no spores oozing out. The sori follow rather closely the circumference of the rotting area and usually develop in rather distinct concentric circles.

The diseased tissue is separated quite sharply from the healthy. At the same time that the rotting is developing outwardly it is also progressing inwardly about as rapidly and a cross section of the apple shows the rot deepest over the center and narrowing from this to the border where it may be quite shallow. The rotting progresses rapidly and there is usually skrinking of the tissues so that the center is more or less depressed. Another peculiarity of the rotten part is its bitter taste, hence the common name of bitter rot. Sometimes other agents aid in the rotting but ordinarily in the fruit attached to the tree the decay is due entirely to the bitter rot fungus. The characteristic appearance of an apple after bitter rot has made considerable headway is shown in Plate C, Fig. 2c. In this case the spores have oozed out in numerous small globules arranged in concentric circles and have become dried up.

Conditions Affecting Development, etc. The effect of moisture on the production of spores was shown in some of the inoculation experiments. Inoculated apples kept unprotected in the air of a room produced numerous fruiting sori but these remained as unruptured black pustules, while similar apples surrounded by a moist atmosphere, by placing them under small bell jars, produced fruiting pustules that oozed out abundant spores in the characteristic pinkish masses. These conditions are illustrated in (c) and (b) of Fig. 2, Plate D. Bitter rot apples that are kept under moist conditions as they become old change the color of the sori to quite dark olive; also under such conditions, especially if the skin of the apple has been ruptured, a growth of dirty olive mycelium is likely to spread over the surface, Plate H. Fig. 1c. These latter

however, are conditions that are not seen on fruit while attached to the tree.

Bitter rot develops rather fast from the first. The rate of development is undoubtedly affected somewhat by the variety attacked. The difference in development on a very unfavorable and on a favorable variety is shown in Plate F; Fig. 1a, showing the unfavorable variety fifteen days after and Fig. 2a or b the favorable variety ten days after inoculation. The state of maturity possibly may have some influence on the rate of rotting though ordinarily the rotting begins in the green apples. Apparently sound ripe apples that were brought in a trunk from southern Illinois when examined four days later showed rotten areas one quarter to one half of an inch in diameter. In the inoculation experiments, rotten areas one to two inches in diameter were produced inside of ten days. Moisture does not seem to increase the rate of rotting though as stated before it affects the character of spore production. In fact the apples kept in the dry air apparently rotted as fast, if not faster, than those left under more moist conditions, Plate H, Fig. 1 a—c, shows the development, during ten days, of rot on apples kept under different conditions of moisture, (a) being kept in open air, (b) under a bell jar, and (c) under a bell jar with moisture.

Species and Varieties infected. Bitter rot has been reported in Illinois only on the apple though it is known to occur elsewhere on the pear, peach, quince, and grape. It was produced however, on these hosts in the laboratory inoculation experiments, except in the case of the quince where only one, a very green specimen, was inoculated. While capable of growing on a great variety of apples it seems to have a preference for certain kinds. Ben Davis and Grimes' Golden which are favorite varieties in southern Illinois are among those that are severely attacked.

That the fungus is much severer on certain varieties was shown in an orchard near Urbana. In this case two trees of the same variety were so badly infected that all of their apples were destroyed. Other varieties immediately surrounding them and whose branches even interlaced escaped with a slow rotting of a few apples, and the other trees of the orchard showed no bitter rot whatever.

Manner of Infection. Rot generally makes its appearance in July and whether it causes considerable or little damage seems to depend largely on the amount of wet weather during the remainder of the season. As has been shown the spores ooze out of the sori much more vigorously under moist conditions, hence in rainy

weather there is undoubtedly furnished a greater supply of these spores for infection, and at the same time increased moisture on the apples favors germination of such of the spores as have been carried to them.

How are the spores carried from the diseased to the sound fruit to produce infection? They are never of a dusty nature so the wind would not be of much aid in their dispersion. When dry they are more or less glued together and when moist form a somewhat sticky mass. It is quite probable that during rains many of the spores are washed over the surface of diseased apples or even carried to the healthy ones. Rain, however, is an agent that would be of little or no use in carrying spores from one tree to another.

It is undoubtedly true that insects are one of the chief agents of dispersal, especially the small pomaceous flies of the genus Drosophila. These flies are produced very abundantly in bitter rot apples under such conditions of moisture as are favorable for the production of spores. That they can serve as dispersing agents was shown in an experiment where some of them were confined in a moist chamber containing bitter rot and sound apples, the latter being sprayed a number of times with water. In a short time the characteristic bitter rot areas appeared scattered over the surface of the sound apples. The appearance of the apple shown in Plate C. Fig. I, indicates that flies may have been the infecting agents in this case. This apple was gathered soon after heavy rains at a time when bitter rot was common.

Life History. The life history of the fungus so far as determined is as follows: The bitter rot begins to appear upon the green fruit about July and gradually spreads, the spreading and consequent damage depending upon the character of the weather. Apples often keep rotting for the remainder of the season and even after they are stored in the fall. During this parasitic stage the only spore form is the summer or Gleosporium one developed as the pinkish pustules so abundant on the rotten areas under favorable conditions. Being thin walled and unprotected such spores are not adapted to live over the winter and develop the disease the next summer.

However, in the fall and succeeding spring on the mummy apples, the fungus, as a saprophyte, gives rise to the permanent or Gnomoniopsis spore stage. The Gleosporium spores that have not been carried away disappear through germination and more or less of a mat of fungus threads covers the apple. Protected by this, perithecia that develop asci with ascospores, which evidently come

to maturity the next summer, are gradually developed in a stroma. These ascospores are shed out of the asci and perithecia when mature and are then scarcely to be distinguished from the Gleosporium spores. No doubt they are carried by the pomaceous flies to the green apples and thus start the disease again for another year.

So far it has not been discovered that the fungus grows on the twigs and by this means carries the fungus over the winter. Very often the infected apples are entirely rotted on the tree and the mycelium can be found somewhat in the fruit stems but no evidence was found that it passed through these into the tissues of the twigs and succeeded in establishing itself there. A more detailed account of the two stages of the fungus is given as follows:

Summer or Gleosporium Stage.

Spores, sori, and mycelium. This is the stage that is parasitic and so is the one that is commonly seen. As explained before, the fruiting pustules are produced near the surface and under favorable conditions ooze out on the surface innumerable spores to form small pinkish masses. When produced under quite moist conditions these spore masses are quite viscid and hence adapted to be carried by insects that come in contact with them in their search for food. The pink color may possibly serve as a means for attracting insects.

As seen under the microscope the spores are colorless. They are typically oblong with rounded ends, but are occasionally slightly curved. They vary in shape, however, from ovate to narrowly oblong and may occasionally have one end pointed. At their center and usually to one side, a hyaline area is to be seen, and, while normally the spores are simple, a septum is often formed here at the time of germination. When first formed the spores have a uniform protoplasmic content, which, however, soon becomes granular. Spores vary considerably in shape and size in the same sorus. While ranging from 10—28 μ in length by 3.5—7 μ in breadth they vary chiefly from 12—16 $\mu \times 4 - 5 \mu$.

The spores are produced terminally from short fertile threads thickly crowded together to form the fruiting sori just beneath the cuticle. See Plate J, Fig. 6. These fertile threads spring from irregular cells compacted into a stroma of greater or less extent. Some of these cells and threads have an olive tint even before the rupture of the cuticle and it is these that give the black color to the immature pustules. By the development of the fertile threads the cuticle is pushed up and, also apparently somewhat corroded, until it finally ruptures over the center of the sorus, and the spores

which are rapidly produced one after another from the tips of the fertile threads ooze out on the surface in pinkish masses. Eventually spore production ceases and the whole of the sorus becomes stromatic tissue which has gradually assumed an olive color. The spores in the meantime have been carried away or if not, those remaining, under moist conditions, eventually disappear through germination.

The mycelium of the fungus runs abundantly through the rotten tissue of the apple and is made up, at least at first, of hyaline threads varying all the way in diameter from 1 to 7 μ. The threads are more or less septate and branched and soon are provided with granular contents. The mycelium is also found slightly in advance of the rotten tissue. Near the surface by crowding between the epidermal cells and their cuticle it gives rise to the scattered stromatic patches that form the sori.

Spore germination. Spores that are fresh germinate more abundantly and vigorously than those that have been dried out for some time. VanTieghem cell cultures of fresh spores in water showed germination beginning inside of three hours. By the end of five hours some of the germ threads may be two or three times the length of the spore. One, two or even three germ tubes may emerge from a spore, coming from either end, or the sides. Usually the spores protrude a germ thread from either end and at the same time a septum is developed at the hyaline area in the center. The germ tubes usually grow into long, slender, slightly branched threads that gradually become septate and empty at their base. They produce a few characteristic Gleosporium spores from the tips of short or elongated undifferentiated branches. A peculiar feature is the production of oval to spherical enlarged cells on the ends of some of the threads and these while spore-like in appearance differ from the ordinary spores not only in shape but also by their deep brown color. They seem to be somewhat of the nature of chlamydospores as they are produced most characteristically in water or cultures with little nutriment. Often they send out a slender hyaline thread as if in germination, and on the end of this produce a second one. A light colored spot, apparently a germ pore, is usually seen at their center. In cultures containing nutriment one sometimes finds gradations between these bodies and the ordinary colored threads and in such cases it may be that they represent the first efforts towards the formation of the permanent stage of the fungus. The various phases of the germination of spores in water are shown in Plate I, Figs. 1–4.

In Van Tieghem cell cultures with potato agar the germination is much more vigorous. Two to several germ threads may be

developed, often two threads from the same end, with the septum formed at center of the spore. These germ threads become longer, much more branched and retain their protoplasmic contents for a longer time though soon becoming septate.

The various stages in the development of a single spore are shown in Figs. 5–11, Plate I. In this case the spore began to germinate about four hours after being placed in the medium and before ten hours had sent out two germ tubes, of which one was more than twice the length of the spore. In less than twenty-four hours it had produced the much branched and septate mycelium partially shown in Fig. 9 and twenty-eight hours from starting was beginning to produce spores. The spores, as in water cultures, are produced terminally on longer or shorter branches and as soon as one is fully grown it falls off and another starts from the same place. It originates as a roundish knob that rapidly elongates into the mature oblong spore. As it develops to its normal size it becomes more and more constricted at its point of attachment until it is entirely cut off and falls away. The rapidity with which these spores can be formed is shown in Figs. 12–19, Plate I, three mature spores being formed from the end of the same branch in about seven hours. Upon germination under unfavorable conditions a bunch of spores sometimes becomes bound together by their short connecting germ tubes, as is shown in Fig. 20 of this same plate. This is a condition similar to that sometimes found in the old olive colored fruiting pustules of apples kept in a moist place, except in that case the spores are usually more or less olive colored.

Artificial cultures. This fungus grows very readily in cultural media and often pure growths may be obtained merely by transferring a few spores from the top of a fresh sorus to the nutrient medium in a test tube. Cultures were made both this way and by the ordinary Petri dish separation method of securing isolated pure colonies from which mycelium was transferred to the test tubes. The media used for cultures were potato agar, apple agar, and apple agar corn meal, and each presented growths somewhat characteristic.

On apple agar slant tubes an evident white growth was visible along the needle streak at the end of the first twenty-four hours. By the end of the second day this growth had increased considerably and spores were being produced rather freely. Usually they were formed from the ends of the flocculent threads without any attempt of grouping into a sorus. Sometimes, however, where

numerous spores have been imbedded in the medium along the needle streak, there were formed more compact masses somewhat of the nature of a sorus—there being found the stromatic layer of irregularly rounded cells giving rise to the densely packed fertile threads about twice the length of the spores produced from their tips. In these cases spores were produced in such numbers as to form pinkish masses. Soon colored threads appear among the hyaline and spore formation decreases. The mycelium spreads over the surface of the agar to form eventually a dense felt of olive colored threads. See Fig. 2 a—b, Plate H. The spores soon disappear through germination and add to this growth. However in those cases where the spores have been produced so abundantly as to form the pinkish masses they remain for some time. The mycelium is made up of branched septate threads from 7 μ to 1 μ in diameter. The olive colored threads are sometimes seen terminating in a hyaline part. These colored threads produce few spores so that with their appearance spore formation diminishes. In their tinted condition the threads usually have more granular contents than when hyaline.

In potato agar cultures the chief difference is that, while the Gleosporium spores are produced somewhat more abundantly, the felt of mycelium on the agar is not so dense and remains white. In time, however, small dark spots may appear in it or sometimes on the edges it becomes somewhat olive tinted. See Fig. 2 d—e, Plate H.

In apple agar corn meal the fungus develops most luxuriantly. At first a white growth is formed extending more or less into the air, but gradually the mycelium penetrates all through the medium. Gleosporium spores are formed very abundantly and often give a pinkish color to the places where produced most luxuriantly. While the mycelium is at first white, as it grows through the medium, it eventually gives this a mottled appearance due to the later production of olive colored threads. In time the Gleosporium spores disappear through germination.

In all of these media there eventually appeared a different stage of the fungus which will be discussed later under the heading of permanent or Gnomoniopsis stage.

Artificial infection. Artificial infection was produced both with picked apples and other fruits and with apples still attached to the trees. The fruit was not sterilized in any way as it was desired to study the disease under natural conditions, as nearly as possible. As the Gleosporium is a very vigorous parasite there was little interference by other agents of decay except in some

cases where ripe fruit was used or where the inoculate fruit was kept in too moist an atmosphere. Most of the experiments were with green apples.

It was found that the bitter rot fungus would develop on a variety of fruits if spores were inserted under the epidermis by means of a needle. In this way there were infected apples, pears, peaches, grapes, and even a green tomato. The only failure was in the case of a very hard, green quince. Infection was also accomplished in many cases merely by placing spores in a drop of water on the unpunctured skin or by means of an atomizer spraying the spores in water over the surface of the fruit.

Inoculation by puncture was usually evident by the end of the second, and by the end of the third or fourth day often had produced a rotten area a quarter to a half an inch in diameter, in the center of which the fruiting sori were appearing more or less abundantly. When the spores were placed on the unpunctured skin, if successful, the rotting usually began at least two or three days later than with the punctured fruit. The difference in the development of rot in punctured and unpunctured apples is shown in (a) and (b) of Fig. 2, Plate D.

The effect of moisture on the production of spores and on the rate of development of the rot has already been described. The following paragraphs show some of the particular results of the chief experiments.

Experiment No. 1549. Two to four green apples each of Duchess, Ben Davis, Maiden Blush, Snow, and Longfield were inoculated while on the trees with spores from a bitter rot apple by placing these under the punctured skin with a scalpel. Examined 41 hours later and found all the apples beginning to rot around the punctures, forming rotten spots about ¼ inch in diameter. At end of 65 hours the rotten areas averaged about ⅓ of an inch and the fruiting sori showed as unruptured black pustules. At end of ten days the rotten areas varied from ⅔ to 1½ inches and all were producing spores though not with equal abundance. The Duchess was the most matured variety and also the worst infected. Apples of two of the varieties photographed ten days after inoculation are shown in plate F., Fig. 2 a–b.

Experiments Nos. 1551–3. Loose green apples were inoculated with bitter rot spore as follows : 1551 with spores in drop of water on unpunctured skin of apple in moist chamber ; 1552, the same except spores were inserted with scalpel beneath the skin ; 1553, the same as 1552 except the apple was left in open air. At end of five days both 1552 and 1553 showed rotten areas about ¾ inch in

diameter and both were producing abundant fruiting pustules but with this difference that on apple kept in open air they showed only as black unruptured sori while on apple kept in moist chamber they had oozed out abundance of spores in the pinkish masses. No. 1551 did not show rot until the seventh day when it had formed a small spot $\frac{1}{16}$ of an inch in diameter. At end of eleventh day it had formed an area $\frac{2}{3}$ of an inch and had numerous unruptured pustules that next day were freely oozing out spores. See Plate D, Fig. 2 for photograph taken on this latter date of the three apples.

Experiment No. 1559. This was an experiment to determine if bitter rot would spread from inoculated apples to others on the same tree. An isolated tree of unknown variety in the old Univiersty orchard was selected and fifty apples punctured, on Aug. 3d, each three times with a needle containing spores. At the end of two weeks the rotten areas were about $\frac{3}{4}$ to 1 inch in diameter, and while a few of the fruiting pustules were producing spores they were chiefly unruptured. On August 22d, as many of the apples had fallen from the tree, 50 to 60 more were inoculated. Sept. 2d found most of the inoculated apples, as well as the others, fallen from the tree due to the very dry weather and the winds. Those first inoculated had rotten areas about $1\frac{3}{4}$ inches in diameter with numerous unruptured sori, while those inoculated later had rotten spots $\frac{1}{4}$ to $\frac{1}{3}$ of an inch without sori. When the experiment first began the weather was dry and very unfavorable for spore production and the spread of the disease, but about August 19th there were good rains. The variety, however, proved to be very resistant to bitter rot, as the rotting developed very slowly and the sori never fruited abundantly, and this together with the falling of the fruit was against the spreading of the disease. The result was that the rot did not spread to a single new place even on the inoculated fruit. Plate F, Fig. 1, shows an apple each of the first, (b) and second (a) inoculation photographed on September 5th.

Experiment No. 1560. In this case three apples each of twelve different varieties were placed in moist chambers after being treated as follows: (a) had spores placed beneath the skin by a scalpel puncture; (b) had spores placed in a drop of water protected from evaporation on the unpunctured skin: (c) was an untreated apple used as a check. On the third day all of the punctured apples showed rotten areas from $\frac{1}{4}$ to $\frac{1}{2}$ of an inch in diameter, but none of the others showed signs of rot. On the fifth day the rotting areas were producing fruiting sori somewhat except in three cases, and on the twelfth day the rotten areas varied

from ¾ to 1⅜ inches. On this day the apples with spores on unpunctured skin showed rotten places ¼ to ⅞ of an inch in four of the varieties, the other eight as well as the checks being sound. The apples were kept under a too damp condition and this produced on their rotten part more or less of a felt of olive colored mycelium. This mycelium came from the punctured places and from the germination of the spores, and in some cases became impure with the growth of other fungi especially with a species of Mycogone. In some cases where this felt had apparently remained uncontaminated there were found, about two months after the infection, signs of a mature stage developing in a stroma beneath it, and in one case there were found mature perithecia with ascospores similar to those produced in the artificial cultures, which are described later on in this paper.

Experiment No. 1590. In this case four varieties of green apples were pricked each ten times around the blossom end with a needle containing spores and then placed under cover. A fifth or check apple was pricked in the same manner with a sterilized needle. Three days later the inoculated apples showed rotten areas $\frac{1}{16}$ to ¼ of an inch around each of the punctures, and two days after this the rotten areas were beginning to merge into one another and were forming spores abundantly in the sori. Plate E shows these and the check apple on this date.

Experiment Nos. 1592–6. These were similar to 1590 but on the following fruits: hard green pear, ripe pear, very hard green quince, hard green peach, soft green peach. At end of three days the inoculations had taken effect at all of the punctures on the pears and peaches except in three cases, forming rotten areas $\frac{1}{16}$ to ½ inch in diameter and rotting most vigorously on the softer pear and peach, especially on the latter which was producing numerous Gleosporium spores. By the end of the eighth day all of the peaches and pears were producing abundance of spores and in the softer ones the rotting had become very extensive. The quince never showed signs of rotting though kept for some time. It evidently was too hard and green, for the most the fungus could do was to form a very slight growth at one or two of the punctures. Quinces in nature, however, are sometimes severely attacked by bitter rot. Plate G, Fig. 1 shows the two peaches on the fifth day after inoculation, the softer one (b), showing sunken areas producing abundance of spores,

Experiment No. 1597. Punctured part of a bunch of green grapes with a needle containing spores of bitter rot and part with a sterilized needle and then placed the bunch in a moist chamber.

A drop of liquid oozed out at each puncture and at end of second day the inoculated ones showed a slight growth of white threads at these places, and in some cases a small rotten area. At the end of a week many of the inoculated grapes appeared as if ripening and some showed numerous but chiefly unruptured sori. Plate G, Fig. 2, photographed thirteen days after puncturing, shows the sound (a) check grapes and the inoculated ones (b) with numerous fruiting sori.

Experiment No. 1614. In this experiment spores were sprayed over the upper surface of a ripe pear, a green peach, a ripe crab apple and a green apple while a check apple was merely sprayed with water. All were kept in a moist chamber and were sprayed several times with water to keep them moist. Four days later the pear was rotting badly but this, apparently due to its ripe condition, was largely caused by saprophytic fungi. The green peach showed on the sprayed surface at least twenty sunken rotten spots, about ¼ of an inch in diameter, the largest of which had sori producing spores. The crab showed very many rotting areas on the sprayed surface, varying from very small to those ⅓ of an inch and often so close together as to merge into one another. The largest of these were producing spores. The green and the check apple developed no rot. Plate D, Fig. 1, shows the condition of the crab apple eight days after being sprayed.

Experiment No. 1637. This was an experiment in the University orchard where eighteen varieties of apples were used. In each case a few apples were inoculated by needle punctures while others merely had the spores sprayed over the surface at the stem end. It was not thought likely that these latter apples would develop the rot as they were sprayed during dry weather, when the water soon evaporated, and in fact none of them became infected. Most of the punctured places took effect and when the apples were picked eighteen days after inoculation they showed rotten areas varying from ¾ to 2 inches in diameter with most of them producing fruiting sori in varying degrees. Among those on which the rot developed most vigorously were Jonathan, Snow, Maiden Blush, Ben Davis, Grimes, Minkler, and Dominie, though the experiment was not extended enough to assert that these were the most susceptible varieties.

Experiment No. 1648 was an attempt to inoculate a green tomato by puncturing several times with a needle containing bitter rot spores. At first it appeared as if this was a failure, as the rot did not start. The tomato, however, began slowly to ripen and later to rot around the punctures, two of which developed fruiting sori of the fungus.

Experiment No. 1584 was one in which the spores of a different Gleosporium—one which was reported as doing considerable damage to Kentia palms in a Chicago greenhouse by killing the leaves from the tip downward and by rotting off the stems at their base—were used to inoculate a green apple. This apple rotted readily and produced numerous sori whose spores were scarcely distinguishable from those of the ordinary bitter rot though the general appearance of the rotted apple was somewhat different.

Permanent or Gnomoniopsis Stage.

Artificial cultures. In practically all of the cultures that were made, including the petri dish separation cultures, there developed in time an ascomycetous fungus that proved to be the permanent stage of the bitter rot. This generally appeared, more or less matured, within two weeks after the cultures were started and usually after the Gleosporium spores had chiefly disappeared through germination. So far as is known this is the first time that the permanent stage has been found. As it appeared so commonly in the cultures it seems rather strange that it was not discovered before, especially by Stoneman* who worked with cultures of this as well as with those of other Gleopsoriums and who described the new genus Gnomoniopsis as the permanent stage of several of these.

In the different media used by the writer some difference in the luxuriance and frequency of this stage was observed. On the apple agar, as has been described under the temporary or Gleosporium stage, the mycelium eventually formed a felt of dark olive threads covering the surface. It is imbedded in this and usually completely concealed by it that the permanent stage appears. Using a needle to pick away the matted threads of the felt one may strike a harder spot which when uncovered shows as a stromatic papilla or cushion slightly elevated above the surface of the agar and about $\frac{1}{16}$ to $\frac{1}{4}$ of an inch in diameter. Usually but few of these were found in a tube. Sections through this stromatic cushion show it to be made up largely of perithecia both on the surface and entirely imbedded in it. These often appear to be in various stages of development and the larger ones are usually more or less united. See Plate J, Fig. 8.

In the potato agar the stromatic cushions are less common and usually smaller. They are also less hidden from view because of the less dense growth that the mycelium makes on this medium

*Stoneman, Bertha. A Comparative Study of the Development of some Anthracnoses. Bot. Gaz. 26:69-120. Aug. 1898. [Illustr.]

and also because they are associated with an evident growth of colored threads that shows as a small black patch in the white mycelium.

In apple agar corn meal tubes the permanent stage developed very vigorously. Soon after the corn meal began to have the mottled appearance described before small black modules appeared that gradually increased into conspicuous stromatic cushions about ¼ of an inch in diameter when fully grown. They developed scattered throughout the medium so that eventually there were to be found old ones at the top whose perithecia had emptied out their spores and lost them through germination, in the center mature ones whose spores had just oozed out in pinkish masses on the surface, and at the bottom those whose perithecia still contained the asci and ascospores. These stromatic cushions developed not only downward but also inward all through the medium. Plate H, Fig. 2 c, shows a photograph of a test tube containing this medium in which a few of these stromatic cushions can be seen.

These cushions arise from the dark olive threads becoming interwoven into quite dense masses forming a sort of false tissue. Sometimes the perithecia look as if they had been fashioned full size out of this. However in sections of a stroma showing all gradations in the formation of the perithecia what appeared to be the youngest condition consisted of a few hyaline somewhat polygonal cells forming a little colony in the dense mat of colored threads. These hyaline cells apparently increased in number, the outer ones becoming tinted and forming the several layers of the perithecial wall (or this in part may arise from the stromatic tissue) while the inner ones continued to fill this differentiated perithecium with delicate, hyaline, polygonal cells. Eventually there appeared toward one side a number of more elongated hyaline cells and these continued to develop at the expense of the polygonal cells, finally forming the full grown asci each with eight ascospores. See Plate J, Fig. 7 for immature stage of perithecia that appeared in nature on an apple.

Sometimes in the cultures the chlamydospore-like bodies described before were seen and these had the color and in the irregular forms were shaped something like the cells of the perithecial wall. Whether they have any special significance in the development of the ascosporic stage is doubtful as one may find all gradations between the quite regular spore-like forms and those that represent only irregular threads.

While mature perithecia were sometimes found in the cultures inside of two weeks usually most of them were not so far advanced by

that time. In fact some of the more imbedded perithecia apparently never developed asci. This condition seemed to be due to the absence of the inner hyaline cells. The stromatic layer when full grown is largely made up of perithecia so that while their primary shape is spherical they often become more or less flattened or irregular by their crowding together. Apparently they developed no especial beak but discharged the spores by a pore or break in the perithecial wall. These oozed out on the surface and in the corn meal cultures were seen as small pinkish globules. With sufficient moisture they soon disappeared through germination. There were no signs of paraphyses. The asci are quite ephemeral soon rupturing and disappearing so that it is difficult to tell an empty perithecium from a sterile one. Because of this temporary nature of the asci and ascospores it is necessary to examine the perithecia at a certain stage in order to find them. The germination of the spores often gives rise to a secondary growth of whitish threads in the cultures.

The size of the perithecia, asci, and ascospores depends somewhat, apparently, on the character of the cultural medium. In general, however, the perithecia were found to vary from 125 to 250 μ in length, while the more or less polygonal reticulations of their walls were about 6—14 μ. The asci were rather numerous in a perithecium, the number probably depending somewhat on its size. Usually they had an oblong or clavate shape tapering somewhat to either end and often with some indication of a pedicle. See Plate J, Fig. 4. In those measured the size varied from 55—70 μ in length, an average size being about 60 x 9 μ. The spores are very similar to those of the Gleosporium stage, in fact when discharged from the asci it is difficult to distinguish the two. See Plate J, Fig. 1 a—b. Their chief difference is that the ascospores are usually slightly curved while the Gleosporium spores are usually straight. The ascospores are not quite so variable in size, ranging from 12—22 μ in length by 3.5—5 μ in width. They are arranged in a biseriate manner in the ascus and are usually, if not always, eight in number. Like the Gleosporium spores, they are colorless and have the central hyaline area.

Germination. The ascospores show no difference in germination from the Gleosporium spores, giving rise to that stage. Like them upon germinating they usually form a septum at the central hyaline area. In water the germination is not very vigorous and but few Gleosporium spores are formed. The characteristic chlamydospore-like bodies are produced. In potato agar there is formed a more luxuriant mycelium with an abundance of Gleosporium spores. See Plate J, Fig. 1—3.

INFECTION OF APPLES. Experiments were tried two different times to infect apples with ascospores both by puncturing the skin and placing spores beneath it and by merely placing spores in a drop of water on the unpunctured skin. In both experiments and under both conditions the characteristic bitter rot of the Gleosporium stage was produced.

On the other hand the ascosporic stage was produced from Gleosporium spores placed on an apple that had been sterilized in a pint jar in an autoclave, the treatment baking the apple and scattering it somewhat over the moist cotton on which it rested. In this case a vigorous growth of dark olive colored threads eventually covered the apple. A number of Gleosporium sori were seen to develop at first and to give rise to pinkish masses of spores. Two months after inoculation the apple and cotton were removed from jar for examination. The mycelium had formed a very dense felt not only on the apple but also over the cotton where parts of the apple or its juice had been carried. Various stages in the development of perithecia were found on the apple but they were chiefly immature, as if the culture had not yet reached its final development. However, a few perithecia were found with characteristic ascospores. All of the Gleosporium spores had entirely disappeared through germination except some few which showed as the uniseptate dark olive bodies more or less connected with one another by short germ threads.

PERMANENT STAGE IN NATURE. In only two cases was the mature permanent stage found on apples. One of these was on an apple that had been inoculated with the Gleosporium spores and being kept in a moist chamber had eventually developed a luxuriant growth of matted dark olive threads on the surface of the rotten area. In the other case it was found on an apple that had rotted in nature from bitter rot but later was kept in a moist place for some time. This also had developed more or less of a matted mycelium on the surface. With bitter rot apples that had laid for some time on the ground, a good many cases were found that seemed to indicate the beginning of this stage. Under the conditions that exist in nature this stage evidently develops slowly and ordinarily could not be expected to be found, at least in any abundance, before May to July of the succeeding year. As the writer has not had the opportunity to hunt for it at this time of the year he cannot state how commonly it occurs in nature.

Apples kept in a damp place change the color of the sori as they become old to quite dark olive. Examination of such sori shows that the dark color is due in part to the olive colored threads

of the stroma and in part to similarly colored spore-like bodies that are scattered among the normal hyaline spores. These have much the appearance of the Gleosporium spores except in their color and in the presence of the septum. They are usually found connected with one another by short germ tubes. It has been shown before that the Gleosporium spores develop a septum at germination and under apparently unfavorable conditions for germination, groups of them are sometimes connected by short germ threads; so it is very probable that these colored spore-like bodies are nothing more than Gleosporium spores that have become colored like the mycelium.

It has also been shown that bitter rot apples kept under moist conditions, especially where the skin has been broken open, usually develop more or less of a mat of olive colored threads over the surface of the rotten area. This apparently comes in part from the germination of the Gleosporium spores and in part from the outgrowth of interior mycelial threads. Eventually all of the Gleosporium spores disappear or show only as the two-celled colored bodies spoken of in the preceding paragraph. See Fig. 7, d, Plate J. The fertile threads of the sori with and after the stoppage of spore production become part of the stromatic layer by the formation of septa dividing them up into cells. These stromatic areas are thus gradually increased in size and become olive colored. From sections cut through old bitter rot apples kept in a moist place it seems probable that the permanent stage is developed in the stromata of the old Gleosporium sori or in those that did not mature to produce this stage and perhaps even in the matted growth of threads that develops entirely on the exterior. This exterior growth of threads at least serves during the winter as a protective covering for the fertile cells beneath it. In nature the stromatic cushions are apparently not so prominent as in artificial cultures and the perithecia are no doubt few in number and never deeply imbedded. Plate J, Fig. 7, shows a cross section of an old bitter rot apple having two well advanced perithecia and apparently a very young one.

Classification. In attempting to classify this permanent stage the writer found considerable difficulty as it did not agree with any of the genera given in Saccardo's Sylloge Fungorum. It seems, however, to come under the new genus described by Stoneman to which reference has already been made. In a few minor characters it does not coincide with the description given for Gnomoniopsis but these are such as might arise from a description based partially on the stage as produced on artificial media or the

insertion of characters not really generic. As the bitter rot fungus is merely the summer stage in the life cycle, it becomes necessary to re-name it under the genus of its permenent stage. As has been shown by Southworth* the bitter rot stage has received a number of names. Probably the species described in 1856 by Berkeley[2] as *Gleosporium fructigenum* was the first name applied to it and this is the one that is now in common use. The proper name then for the fungus as now understood is *Gnomoniopsis fructigena* (Berk.)

SCIENTIFIC DESCRIPTION. The generic characters of Gnomoniopsis, as modified somewhat by the writer's investigations, together with the description of the species causing bitter rot may be given as follows:

GNOMONIOPSIS, Stonem. Perithecia membranaceous, dark brown, spherical to flask shaped, often rostrate, sometimes evidently hairy, cæspitose or more or less compound and immersed in a stroma with which they often form an evident hard cushion; asci oblong to clavate, often fugacious, aparaphysate; ascospores hyaline, apparently eight, distichous, oblong, usually slightly curved, unicellular. Permanent stage of Gleosporium-like fungi.

Gnomoniopsis fructigena (Berk.) Clint. n. n. permanent stage developing on decayed pomaceous fruits; forming stromatic cushions (often concealed by dark olive mycelial felt), which contain immersed and more or less compounded, subspherical perithecia; asci subclavate, often slightly pedicellate, fugacious; 55—70 μ in length, ascospores alantoid, with evident central hyaline area chiefly 12—22 x 3.5—5 μ. Gleosporium stage causing rotting of pomaceous fruits; sori small, developing more or less in concentric circles, usually soon rupturing and oozing out spores in small pinkish masses; spores hyaline, chiefly oblong, unicellular, with evident hyaline areas when fresh, 10—28 x 3.5--7 μ but chiefly 12—16 by 4—5 μ.

*SOUTHWORTH, E. A. Ripe Rot of Grapes and Apples. Journ. Myc. **6**:164-73. 1891.

[2]BERKELEY, M. J. *Gleosporium fructigenum*, n. s. Gard. Chron. **1856**: 245.

EXPLANATION OF ILLUSTRATIONS.

Plate A, Fig. 1. Apple showing sun burn.

Plate A, Fig. 2. Apples showing rot started at injured places produced by birds and worms.

Plate B, Fig. 1. Apples showing fruit blotch. (a–b) in the earlier, and (c) in the later stage.

Plate B. Fig. 2. Apples showing stages (a–d) in the development of black rot, (a) not yet having formed fruiting conceptacles, (d) with abundance of them containing mature spores.

Plate C, Fig. 1. Apple showing very young state of bitter rot, with many colonies scattered over its surface.

Plate C, Fig. 2. Apples showing different stages of bitter rot in which colonies have attained considerable size and have developed numerous fruiting sori; (a, c) developed from single and (b) from several colonies.

Plate D, Fig. 1. Artificial infection of bitter rot produced on ripe crab apple, kept in moist chamber, by spraying with water containing spores of bitter rot. Sprayed Aug. 23d and photographed Aug. 31st.

Plate D, Fig. 2. Artificial infection of three green apples under different conditions: (a) spores placed in drop of water on the unpunctured surface and apple kept in moist chamber; (b) spores placed under the skin by needle puncture and apple kept in moist chamber; (c) same as (b) but apple kept in open air; (a) and (b) have numerous sori producing spores, while in (c) they remained as unruptured pustules. Apples treated July 25th and photographed Aug. 6th.

Plate E. Four varieties of green apples (a–d) each punctured ten times around the blossom end with a needle containing bitter rot spores and kept in a damp chamber; (e) check apple merely punctured with sterilized needle. Apples punctured Aug. 16th and photographed Aug. 21st.

Plate F, Fig. 1. Apples of an unknown variety inoculated with bitter rot, by needle punctures. while on the tree; (b) inoculated Aug. 3d, and (a) Aug. 22d, both being photographed Sept. 5th. This proved to be a variety very resistant to bitter rot; (a) developed no sori while in (b) they were chiefly immature.

Plate F, Fig. 2. Apples inoculated with bitter rot, by scalpel puncture, while still on the tree; photographed ten days after inoculation. These two varieties were quite susceptible to bitter rot; they show the fruiting sori and the central sunken areas.

Plate G. Fig. 1. Hard (a) and soft (b) green peach punctured ten times with a needle containing spores from bitter rot apples and then kept in a moist chamber, (b) producing abundance of spores from the fruiting sori. Punctured Aug. 16th and photographed Aug. 21st.

Plate G, Fig. 2. Green grapes (b) each punctured once with a needle containing spores from bitter rot apple and then placed in moist chamber; (a) check grapes, merely punctured with a sterilized needle. Punctured Aug. 17th and photographed Aug. 30th, at which time inoculated grapes were covered with sori producingspores.

Plate H. Fig. 1. Apples showing development of bitter rot under different conditions of moisture; (a) kept in open air, fruiting pustules remaining unruptured; (b) kept under a bell jar, fruiting pustules producing abundance of spores; (c) kept under bell jar with moisture, fruiting pustules producing spores and felt of mycelium growing over the surface. Pin heads mark the boundary of the rot when treatment began on July 26th; photographed Aug. 6th.

Plate H, Fig. 2. Test tube cultures of the bitter rot fungus on different media; (a) front and (b) back view of culture on apple agar, the fungus making a dark growth on this medium; (c) culture in apple agar corn meal, showing a few stromatic cushions of the permanent stage; (d) back and (e) front view of culture on potato agar, the mycelium forming a white growth on this medium.

Plate I. Bitter rot: (a) spores; (b) germ threads on mycelium; (c) chlamydospore-like bodies.

Fig. 1–4, showing germination of bitter rot spores in water in Van Tieghem cells. Fig. 1, condition of spores when first placed in water, 11 a. m., Aug. 30th. Fig. 2, various stages of spore germination at 4 p. m., Aug. 30th. Fig. 3, stages of germination at 10 a. m., Aug. 31st; 3′, showing full length of germ threads of two spores less highly magnified. Fig. 4, general condition of the mycelium 10 a. m.. Sept. 2d, producing chlamydospore-like bodies (c) and a few Gleosporium spores (a).

Fig. 5–11, showing germination of a single spore (5) in potato agar in Van Tieghem cell. Fig. 5, condition of spore 10:30 a. m., Aug. 16th when spore was first placed in potato agar. Fig. 6, 3 p. m., just after spore began to germinate. Fig. 7, condition at 5:30 p. m., Fig. 8, condition at 8 p. m. Fig. 9, condition at 9 a. m., Aug. 17th, less highly magnified to show extent of mycelium at this time. Fig. 10, central part of mycelium of Fig. 9. Fig. 11, partial view of mycelium at 2:30 p. m., showing formation of Gleosporium spores.

Fig. 12–19, showing manner and rate of production of Gleosporium spores in potato agar in Van Tieghem cells. three spores being formed in less than eight hours. Fig. 12, condition at 9 a. m.; Fig. 13, at 9:05 a. m.; Fig. 14, at 10:30 a. m.; Fig. 15, at 11 a. m.; Fig. 16, at 12 N; Fig. 17, at 3:00 p. m.; Fig. 18, 3:30 p. m.; Fig. 19, at 4:10 p. m.

Fig. 20, showing spores that had been produced in Van Tieghem cell uniting by short germ tubes after manner so common with spores in old sori on the apples kept in moist place.

Plate J, Fig. 1. Ascospores from permanent stage of bitter rot fungus; (a) typical spores; (b) spores of somewhat unusual shapes; (c) germinating spores after 16 hours in water in Van Tieghem cell; (d) germinating spores at end of 40 hours.

Fig. 2–3, germination of the ascospores in potato agar; (a) spores; (b) germ threads. Fig. 2, germination at end of 16 hours, a′ being less highly magnified. Fig. 3, part of mycelium showing spore production at end of 41 hours.

Fig. 4, showing two asci with ascospores of the permanent stage of bitter rot, also one free ascospore. Spores somewhat displaced out of their distichous arrangement.

Fig. 5. showing rather diagrammatic drawing of a section through a young bitter rot fruiting sorus on apple; (a) parenchyma cells of apple; (b) cuticle of apple; (c) subhymenial layer of fungus cells; (d) fertile threads of fungus; (e) spores.

Fig. 6, showing character of fertile cells of sorus with attached Gleosporium spores.

Fig. 7, showing section through surface of an old bitter rot apple on which the permanent stage is developing; (a) parenchyma cells; (b) cuticle; (c) fungous threads of the felt; (d) old, dark colored, uniseptate Gleosporium spores. Two perithecia well advanced, one showing young asci starting at its base; also a very young stage of a perithecium.

Fig. 8, showing diagrammatic drawing of a section through part of a stromatic cushion of the permanent stage of bitter rot. This is from a culture grown on apple agar. It shows various sizes of the perithecia (a).

Fig. 5, 7, 8 less highly magnified than the other drawings.

Plate A

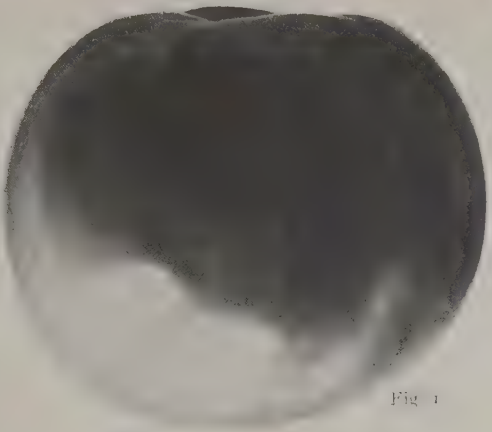


Fig 1

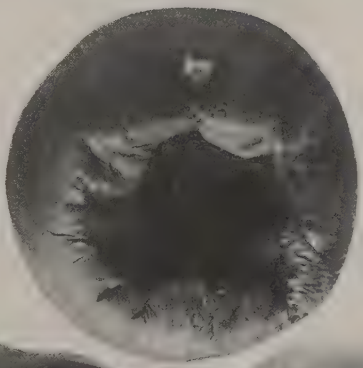


Fig 2

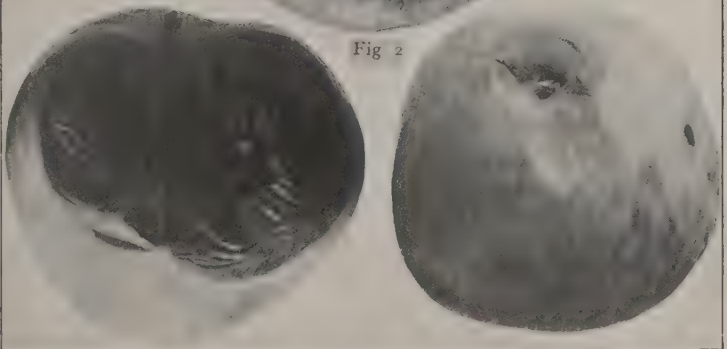


Plate B.
a.
b.
Fig. 1
c.
a.
b.
Fig. 2
c.
d.

Plate C.

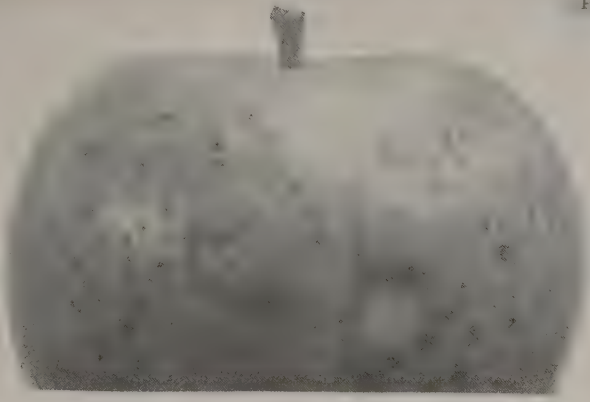
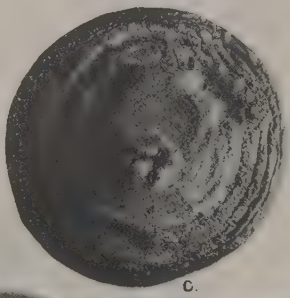
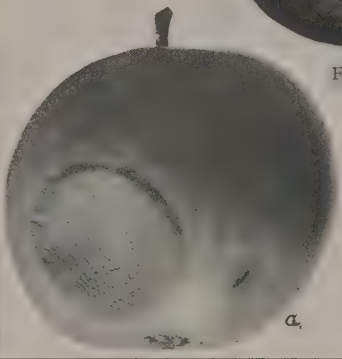


Fig. 1





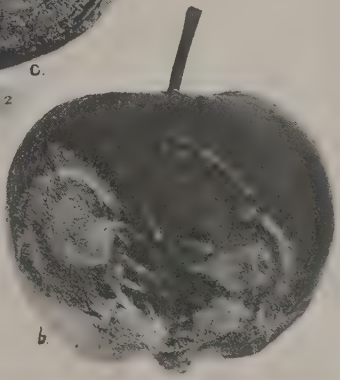


Fig. 2

Plate D.

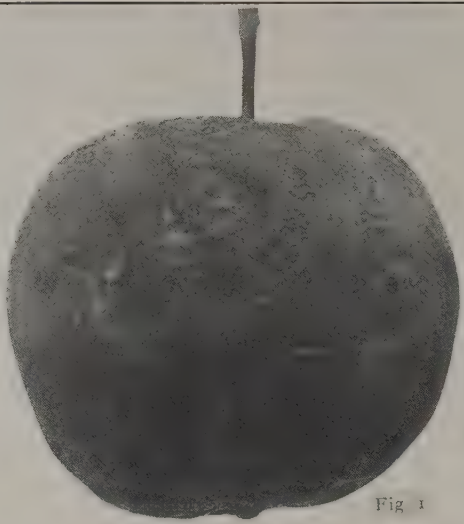
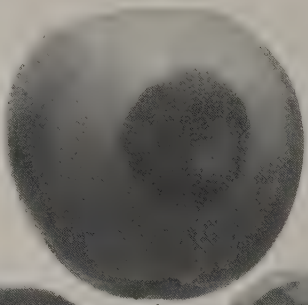
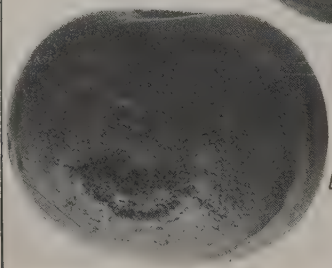


Fig. 1



a



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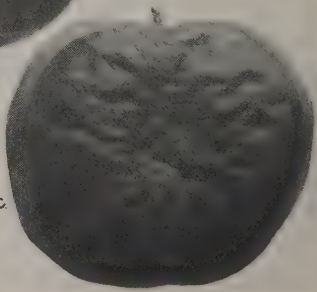


c.

Fig. 2

Plate E.

Plate F

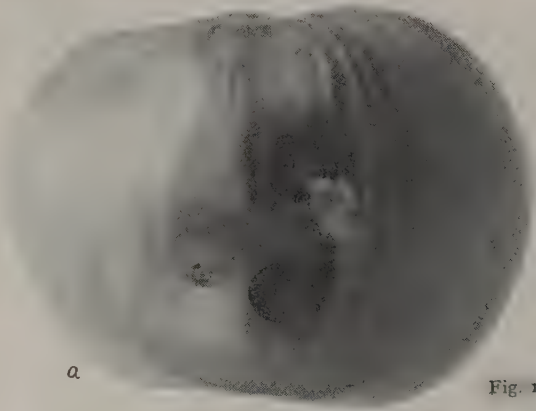
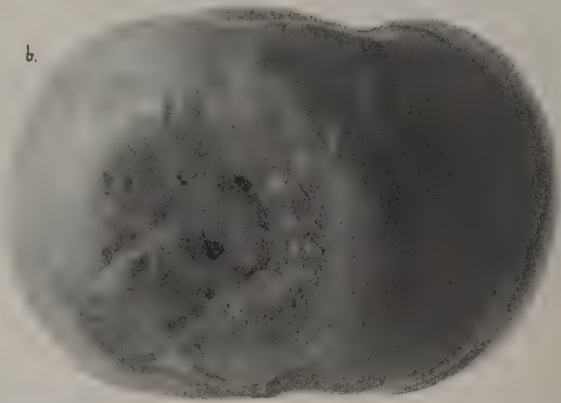


Fig. 1



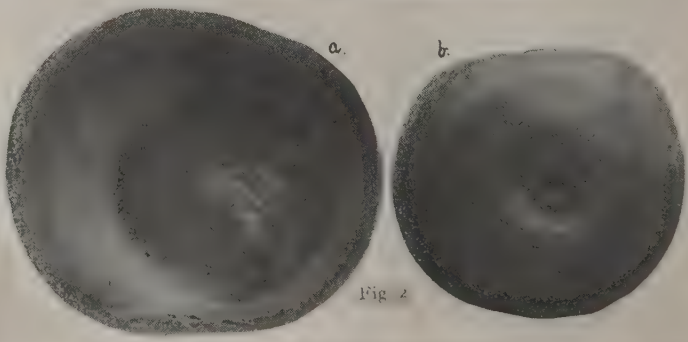


Fig. 2

Plate G.

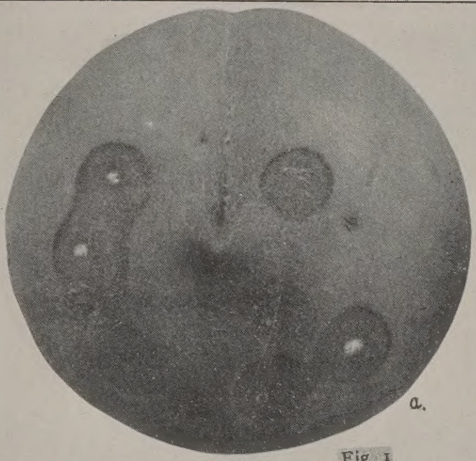


Fig. 1

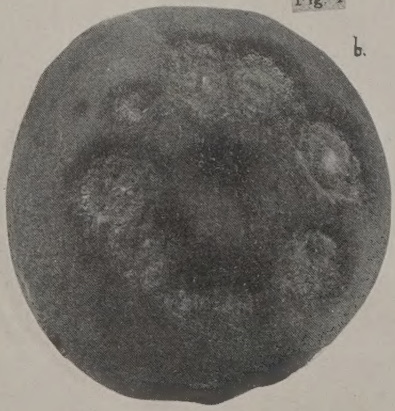


Fig. 2

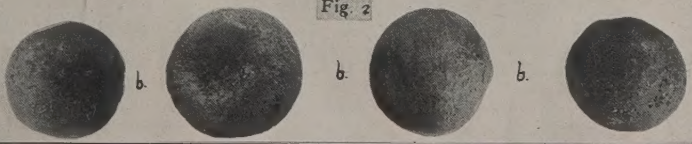


Plate H

Fig. 1

Fig. 2

PLATE I.

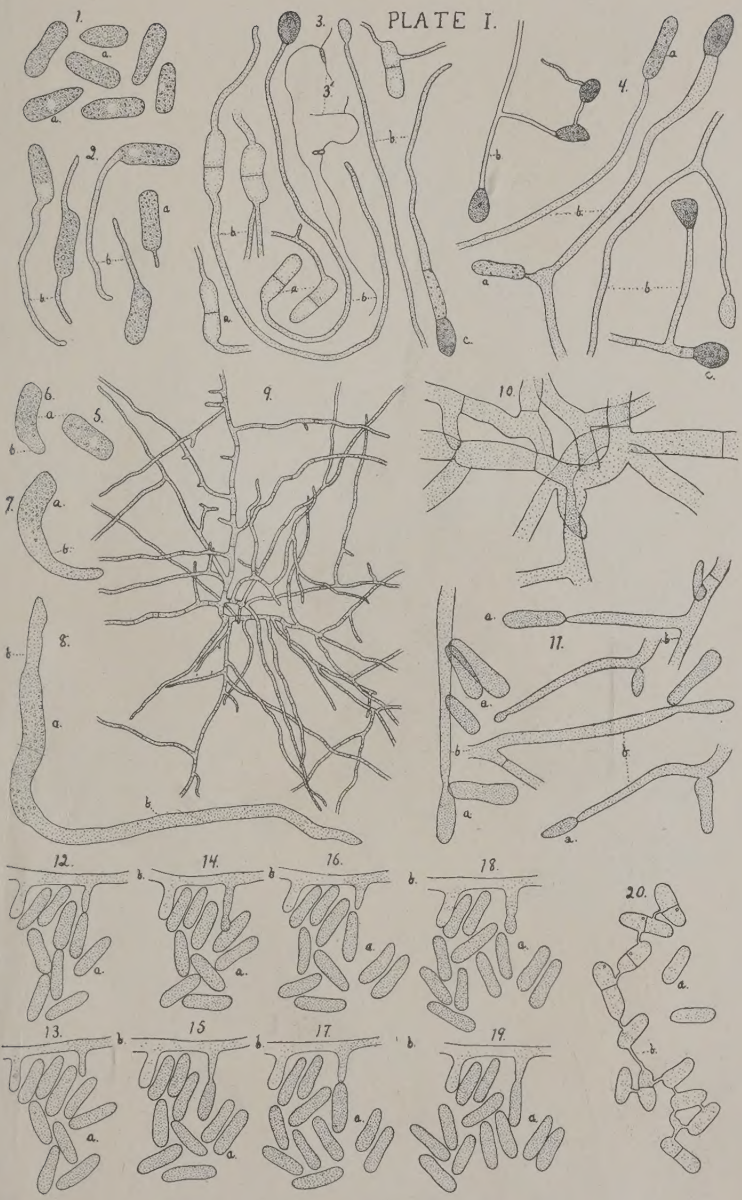


PLATE J.